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> d his

     (FILE 'HOME' ENTERED AT 09:40:34 ON 04 APR 2004)

     FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE' ENTERED AT 09:40:46 ON 04 APR 2004

     FILE 'REGISTRY' ENTERED AT 09:41:35 ON 04 APR 2004
                E CACAATTAAAACTGTGCATTAC/SQEN 5
L1             9 S E3 OR E4

     FILE 'CAPLUS' ENTERED AT 09:42:49 ON 04 APR 2004
L2             4 S L1

     FILE 'REGISTRY' ENTERED AT 09:49:12 ON 04 APR 2004
                E GTAATGCACAGTTTTAATTGTG/SQEN

     FILE 'CAPLUS' ENTERED AT 09:49:12 ON 04 APR 2004
L3             QUE GTAATGCACAGTTTTAATTGTG|CACAATTAAAACTGTGCATTAC
               S L3/SQSN

     FILE 'REGISTRY' ENTERED AT 09:50:14 ON 04 APR 2004
L4          6555 S L3/SQSN

     FILE 'CAPLUS' ENTERED AT 09:50:38 ON 04 APR 2004
L5           141 S L4

     FILE 'CAPLUS' ENTERED AT 09:50:52 ON 04 APR 2004
L6           141 S L5
L7            74 S L6 AND PY<2000
L8             0 S L5 AND SQL<100

=>
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L3 ANSWER 108 OF 123 MEDLINE on STN DUPLICATE 84
ACCESSION NUMBER: 96187329 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8609204
TITLE: Detection of southern African human immunodeficiency virus type 1 subtypes by polymerase chain reaction: evaluation of different primer pairs and conditions.
AUTHOR: Engelbrecht S; van Rensburg E J
CORPORATE SOURCE: Department of Medical Virology, University of Stellenbosch, Tygerberg, South Africa.
SOURCE: Journal of virological methods, (1995 Nov) 55 (3) 391-400.
Journal code: 8005839. ISSN: 0166-0934.
Report No.: PIP-115318; POP-00256126.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Population; AIDS
ENTRY MONTH: 199605
ENTRY DATE: Entered STN: 19960605
Last Updated on STN: 20021101
Entered Medline: 19960524

AB The purpose of the study was to develop a **specific** and sensitive **PCR** protocol using env, gag and LTR **primer** pairs to detect **HIV**-1 **subtypes** present in the Western Cape, South Africa. Twenty-two virus strains, belonging to **HIV**-1 **subtypes** B, C and D, were randomly selected for **PCR** evaluation. Cell lysates prepared from these virus-infected cultured cells were tested using 5 different **primer** pairs: gag SK38/SK39; gag 22/SK39; gag a/b, gag c/d (nested); env SK68/SK69 and LTR SK29/SK30. Eight different **PCR** profiles were evaluated: one profile each for the 3 gag **primer** pairs, 3 profiles for the env and 2 profiles for the LTR **primer** pairs. The number of **PCR** cycles, time per cycle and/or annealing temperature were changed in each profile. The optimum **PCR** profile for a **specific** **primer** pair was defined as that which detected one copy of proviral plasmid DNA after dot-blot hybridisation. Gag **primer** pairs detected **HIV**-1 DNA in all 22 samples. With the env **primer** pair, suboptimal conditions failed to detect most of the **HIV**-1 **subtype** C samples. By increasing the number of cycles and time per cycle, a 100% sensitivity was achieved. With the LTR **primer** pair all samples were detected by decreasing the annealing temperature and increasing the individual cycle times. This confirms that once **PCR** conditions are optimised, all **HIV**-1 **subtypes** in our study could be detected using different **PCR** **primer** pairs. During 1984-92, in South Africa, virologists isolated **HIV**-1 from **HIV**/AIDS patients at hospitals in the Western Cape. Two virologists from the University of Stellenbosch Hospital in Tygerberg selected 22 virus strains, belonging to **HIV**-1 **subtypes** B, C, and D, to study in order to develop a **specific** and sensitive polymerase chain reaction (**PCR**) protocol using env, gag, and LTR **primers**. They used five different **primer** pairs to prepare cell lysates from the **HIV**-infected cultured cells: gag SK38/SK39, gag 22/SK39, gag a/b, gag c/d (nested), env SK68/SK69, and LTR SK29/SK30. The virologists evaluated eight different **PCR** profiles: one profile each for the three gag **primer** pairs, three profiles for the env, and two profiles for the LTR **primer** pairs. They changed the number of **PCR** cycles, time per cycle, and/or annealing temperature in each profile. The **PCR** profile for a **specific** **primer** pair that detected one copy of proviral plasmid DNA after dot-blot hybridization was considered the optimum **PCR** profile. Gag **primer** pairs detected **HIV**-1 DNA in all 22 samples. The env **primer** pair did not detect most **HIV**-1 **subtype** C samples. When the researchers increased the number of

cycles and time per cycle, the env **primer** pair achieved 100% sensitivity. When they decreased the annealing temperature and increased the individual cycle times, the LTR **primer** pairs detected all samples. These findings support that optimization of a **PCR** assay is necessary to achieve high assay sensitivity, specificity, and reproducibility and that **PCR** sensitivity should be considered seriously when interpreting **PCR** results for **HIV** diagnosis.